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# Age-Related Patterns in Urinary Biomarkers Using Urine Dipstick Analysis

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**Abstract:** The urinary system's primary functions are to eliminate waste from the body, regulate blood volume and pressure, control electrolyte and metabolite levels, and maintain blood pH. Aging significantly impacts the urinary system, often reducing the efficiency of its organs. As individuals age, the urinary tract undergoes various changes that can affect its function and are reflected in urine diagnostic parameters. In a study conducted with 100 individuals in Durres, Albania, the sample consisted of 46% women and 56% men. The results revealed that 15% tested positive for proteinuria, 45% for ketones, 5% for high glucose levels, and 6% for nitrites. Statistical analysis indicated a significant relationship between high glucose levels in urine and age ( $\chi^2 = 0.203^*$ , p = 0.043), suggesting that glucose levels may increase with age. Additionally, there was a negative relationship between urine pH and age (p < 0.001), indicating that pH levels tend to decrease as people get older. These findings underscore the importance of monitoring urine diagnostic parameters in aging populations. Urine reagent strips proved to be a valuable tool in this study, offering quick and efficient results and allowing for the detection of a wide range of chemical parameters. This approach provides essential insights into the functional changes occurring in the urinary system with age.

Key words: Urinary system, diagnostic parameters, urine analysis, aging.

# Introduction

The urinary tract functions as the body's filtration system, removing urine, which consists of waste and excess fluid. For normal urination, all components of the urinary tract must work together in the proper sequence. Additionally, the urinary system plays a crucial role in regulating the volume, acidity, salt concentration, and chemical composition of blood, lymph, and other body fluids.

Aging significantly affects the urinary system, as the efficiency of its organs diminishes over time. The population of individuals over 65 is growing rapidly in developed countries, leading to an increase in elderly patients diagnosed with kidney damage and nephrosclerosis (O'Sullivan *et al.*, 2017). Excess protein in the urine indicates that the kidney filters, known as glomeruli, are not functioning correctly and are allowing protein to leak into the urine. Damage to the glomeruli results in conditions such as nephritis or glomerulonephritis, which become more likely with age (Sureshkumar *et al.*, 2003).

Urinalysis is a diagnostic test that examines the visual, chemical, and microscopic characteristics of urine. It may involve a series of tests to detect and measure various compounds in a single urine sample. This test is essential for assessing kidney function, identifying potential infections, and monitoring chronic conditions. Healthcare providers commonly use urinalysis to assess and monitor conditions like liver disease, kidney disease, and diabetes, as well as to diagnose urinary tract infections (UTIs). The urine reagent strip test, or dipstick test, is used to evaluate the chemical properties of a urine sample. Lab technicians use dipsticks with chemical pads that change color when exposed to specific substances (Kutter 2000). Regular urinalysis can be a valuable tool in preventive healthcare, allowing for the early detection of abnormalities before they develop into more serious conditions. By analyzing changes in urine composition, healthcare professionals can gain insights into an individual's overall health and tailor treatment strategies accordingly.

# **Material and Methods**

In this study, urine samples from 100 individuals (both women and men aged 15 to 90 years recommended by the doctor for urine analysis) from the city of Durres, Albania, were analyzed. The samples were collected and analyzed from April to May 2023 at the "Medical Care Lami" medical laboratory in Durrës, in collaboration with the Department of Biology, Faculty of Natural Sciences,

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University of Tirana. The entire procedure adhered to the EFLM European Urinalysis Guideline 2023 (Kouri *et al.*, 2023).

Participants were instructed to collect a urine sample, preferably the first morning urine, in a sterile container. They were advised to avoid alcohol, coffee, tea, smoking, and vigorous exercise for 24 hours prior to sample collection. This protocol was designed to minimize potential confounding factors that could affect the accuracy of the results. After the urine samples were delivered, the necessary analyses were performed.

Urine samples were analyzed using Labstrip U11 Plus test strips (77 Elektronika), which measure 11 parameters: bilirubin, urobilinogen, ketones, ascorbic acid, glucose, protein, blood, pH, nitrite, leukocytes, and specific gravity. For reading the Labstrip U11 Plus urinalysis strips, the DocUReader 2 Pro analyzer was used. Although visual interpretation using a color chart is an alternative method, the DocUReader 2 Pro was employed in this study to ensure higher accuracy. This choice was made to reduce the potential for human error and improve the reliability of the results.

The data on urinary chemical parameters obtained from the Labstrip U11 Plus tests were processed using IBM SPSS (Statistical Package for Social Sciences) version 16 for Windows. To assess the relationship between urinary parameters and age, the chi-square test and Pearson correlation test were used, with a significance level set at p < 0.05. These statistical methods were chosen to identify any significant trends or correlations between age and the various urinary parameters measured.

# **Results**

In our sample of 100 individuals, 46 are women and 54 are men. The age distribution reveals that 47% of the participants are under the age of 40, while 53% are over 40. Notably, 53% of the sample are over this age range where there is a claimed higher probability of abnormalities such as proteinuria due to impaired kidney filtration. To investigate this, we grouped the data into five distinct age categories, as illustrated in Table 1. Our analysis shows that 15% of the individuals tested positive for proteinuria, with the highest prevalence observed in the age group over 73 years.

We applied the Pearson correlation test to assess the linear relationship between age and the presence of proteins in the urine. The test results yielded a p-value of 0.096, indicating that there is no statistically significant correlation between age and proteinuria at the 0.05 significance level. This indicates that although proteinuria appears more frequently in older age groups, the association between age and the occurrence of proteinuria is not statistically significant in our sample. Further studies with larger sample sizes might be needed to explore this potential association more thoroughly.

**Table 1.** Variation in proteinuria levels among different age groups

Age groups (years)	N (%) Normal level of proteins <20 mg/dL	N (%) Abnormal level of proteins >20 mg/dL	Total
<28	25 (86.2)	4 (13.8)	29
29-43	19 (95.0)	1 (5.0)	20
44-58	18 (90.0)	2 (10.0)	20
59-73	16 (84.2)	3 (15.8)	19
>73	7 (58.3)	5 (41.7)	12
Total	85	15	100

Urine pH changes with increasing age, making it an important parameter to assess in this study. Table 2 illustrates the distribution of urine pH across different age groups of the participants. It is evident from the data that older individuals tend to have lower pH levels compared to their younger counterparts.

Specifically, 57% of the sample exhibited a pH of 5, indicating an acidic environment. In contrast, 30% of the individuals had a pH in the range of 5.5-6, which is generally considered the optimal pH level for urinary health. The prevalence of acidic pH in a significant portion of the sample suggests that age-related changes in urine pH could be a noteworthy factor in assessing overall health.

To explore the relationship between age and pH, a Pearson correlation test was conducted. The results confirmed a significant negative correlation between age and urine pH (p < 0.001). This finding indicates that as age increases, urine pH tends to decrease, meaning older individuals are more likely

to have lower (more acidic) pH levels.

These results underscore the importance of considering urine pH in conjunction with age when evaluating health status. Lower urine pH may be associated with various health conditions or metabolic changes, making it crucial for healthcare providers to integrate this information with other clinical data to make informed decisions. Regular monitoring and assessment of urine pH could be beneficial in identifying potential health issues early and managing them effectively.

**Table 2**. Variation in urine pH among various age groups

	pН					
Age groups (years)	5.0	5.5	6.0	6.5	7.0	Total
<28	8	8	3	5	5	29
29-43	10	4	4	2	0	20
44-58	17	0	3	0	0	20
59-73	16	2	1	0	0	19
>73	6	3	2	1	0	12
Total	57	17	13	8	5	100

Table 3 below illustrates the distribution of ketones levels across different age groups of the participants. From the results obtained, it is noted that 45% of the individuals of this sample are positive in terms of the high presence of ketones in the urine, while 55% of the individuals are negative. Chi-square test was used to test whether there is a relationship between ketones and age. From the data we obtained, it turned out that there is no significant relationship between ketones and age (p= 0.068,  $\alpha$ =0.05).

**Table 3**. Variation in ketones levels among various age groups

Age groups (years)	N (%) Normal level of ketones <5 mg/dL	N (%)Abnormal level of ketones >5 mg/dL	Total
<28	12 (41.4)	17 (58.6)	29
29-43	11 (55.0)	9 (45.0)	20
44-58	13 (65.0)	7 (35.0)	20
59-73	9 (47.4)	10 (52.6)	19
>73	10 (83.3)	2 (16.7)	12
Total	55	45	100

Figure 1 illustrates the distribution of glucose levels within the study sample. Out of 100 individuals, 95 exhibit normal glucose values, while 5 individuals (5% of the sample) have elevated glucose levels. To analyze the association between glucose levels and age, a Chi-square test was conducted. The results reveal a Chi-square statistic ( $\chi^2$ ) of 0.203, with a p-value of 0.043. Given the significance level ( $\alpha$ ) of 0.05, the p-value is less than  $\alpha$ , indicating a statistically significant relationship between glucose levels and age. The significant p-value (0.043) suggests that there is a meaningful relationship between glucose levels and age in this sample. This finding implies that age is likely associated with variations in glucose levels, highlighting the importance of considering age as a factor in glucose level assessments and potentially guiding targeted interventions for individuals based on their age group.

Table 4 below illustrates the distribution of nitrinuria cases across different age groups of the participants, when 94% of individuals in the sample were negative and 6% positive. Any degree of pink color in the urine is interpreted as a positive nitrite test representing the presence of 105 or more organisms per milliliter. Using the Pearson correlation test, we analyzed whether there is a significant relationship between nitrites and the age of individuals. From the data obtained, it appears that there is no relationship between them (p=0.668).

The specific gravity (SG) in this sample has a range of normal values, we have a minimum value of 1015 and a maximum value of 1035 (Figure 2). The value 1030 has the highest frequency, with 46

individuals out of 100 total. To test whether between age and SG the Pearson correlation test was used, p=0.325. From this data we conclude that there is no relationship between these two variables. All other parameters in our sample of 100 individuals, which were tested with LabStrip U11 including bilirubin, urobilinogen, ascorbic acid, blood and leukocytes, were normal (for urobilinogen) or negative (for bilirubin, ascorbic acid, blood and leukocytes).

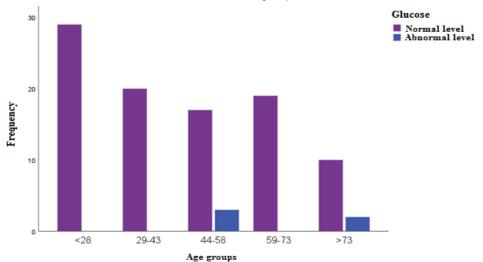


Figure 1. Variation in glucose levels among different age groups

Table 4: Prevalence of Nitrinuria Across Various Age Groups

Age groups (years)	N (%) Negative for nitrites	N (%) Positive for nitrites	Total
<28	27 (93.1)	2 (6.9)	29
29-43	18 (90.0)	2 (10.0)	20
44-58	20 (100.0)	0 (0.0)	20
59-73	19 (100.0)	0 (0.0)	19
>73	10 (83.3)	2 (16.7)	12
Total	94	6	100

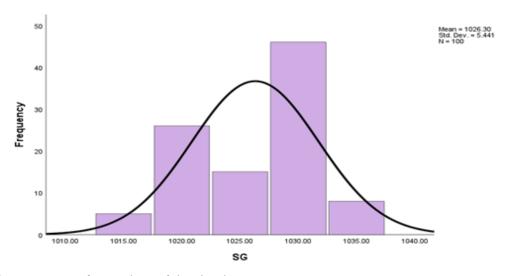


Figure 2. Frequency of SG values of the simple

# **Discussion and Conclusion**

In our study, the presence of elevated protein levels in urine was found in 15% of cases. This aligns with the range reported in other studies, which varies from 14.1% to 24.9% (Parker *et al.*, 2020; Rosenstock *et al.*, 2018). Proteinuria, or the presence of abnormal amounts of protein in urine, serves as an early indicator of chronic kidney disease (CKD). The prevalence of CKD in Europe is reported

to range from 3.3% to 17.3% (Brück *et al.*, 2016). While CKD can occur with normal protein levels in urine, proteinuria can also be an early sign of other conditions such as diabetes, hypertension, and cardiovascular diseases.

Our study also identified a negative correlation between urinary pH levels and age, indicating that as individuals age, urine pH tends to decrease, becoming more acidic. Highly acidic urine can result from several conditions including acidosis, uncontrolled diabetes, diarrhea, starvation, dehydration, and respiratory diseases associated with carbon dioxide retention and acidosis. Some studies suggest that low urine pH might signal chronic kidney disease, heart failure, metabolic abnormalities, or other disorders (Nakanishi *et al.*, 2012; Kraut & Madias, 2016).

In our sample, 45% of individuals exhibited high levels of ketones in urine. Ketoneuria can be a useful screening tool to estimate the ketonic state of the body. Elevated ketone levels may indicate a risk of ketoacidosis. Factors contributing to elevated ketone levels include prolonged fasting, a low carbohydrate diet, certain medications, excessive alcohol intake, and dehydration. Studies have associated ketone presence in urine with metabolic health and weight (Kim *et al.*, 2020) and with aerobic exercise (Han & Jeo, 2020).

Glucose levels in urine are considered normal in small amounts. However, glucose concentrations exceeding 25 mg/dL in random fresh urine samples are indicative of a pathological condition known as glucosuria. In our sample, 3% exhibited glucosuria. A significant relationship was observed between high glucose levels in urine and age (p=0.043). Elevated glucose in urine may suggest conditions such as diabetes, gestational diabetes, or certain kidney disorders, which are more prevalent in older adults (Kalyani & Egan, 2013).

The presence of nitrites in urine, found in 6% of our sample, is generally associated with bacterial infections in the urinary tract. Nitrite testing is based on the ability to convert nitrate to nitrite, with production linked to certain bacteria such as members of the Enterobacteriaceae family. It is important to note that not all bacteria reduce nitrates, so a negative test does not rule out infection (Franz & Harl, 1999).

Specific gravity (SG) measures the kidneys' ability to regulate water content and excrete waste. It plays a crucial role in diagnosing conditions affecting urine water content. SG values can vary based on fluid intake: a low SG (around 1.001) may be normal with high water consumption, while a high SG (above 1.030) may be normal with low fluid intake. Insufficient water intake has been linked to health conditions such as renal insufficiency and cardiovascular issues (Clark, 2011; Dmitrieva, 2022).

Diagnosing urinary parameters is crucial for identifying potential disorders based on abnormal values. Abnormal findings may indicate various conditions, some of which could be serious. It is essential to study these parameters in conjunction with physiological mechanisms and overall kidney function. The results from such analyses should assist in differentiating between normal samples and those requiring further examination for possible renal or genitourinary tract disorders. Positive samples may warrant additional investigations such as microscopic examination, chemical analysis, or bacteriological tests (Oyaert & Delanghe, 2019).

Urine specimen analysis serves as a rapid, first-line screening method in a multi-test workflow. Due to its wide availability and ease of use, it provides clinicians and patients with prompt point-of-care information. While visual inspection of urine samples might be subject to subjective color interpretation, automated analyzers offer more reliable results (White *et al.*, 2011; Rowell, 1998).

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# Human Health Risk Prospecting Against Lead, Chromium, and Cadmium in Consumed Herbal Snuffs in Sokoto State, Nigeria

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**Abstract:** Nowadays, there is much attention on the utilization of herbal medicines among many parts of the world including Sokoto. However, there are concerns that the current trend in pollution could lead to an uptake of lead, cadmium and chromium by herbal plants and in turn can harm the consumers. Therefore, the objective of this paper was to assess cadmium, chromium, and lead in some herbal snuffs consumed in Sokoto, Nigeria and estimate human health risk. Standard methods and materials of analytical grade were used for this work. The assessed concentrations (p < 0.05) of Cd, Cr, and Pb in some common herbal snuffs consumed in Sokoto, Nigeria show ranges of Cd, Cr, and Pb respectively as follows:  $0.07 \pm 0.001$  to  $0.13 \pm 0.005$  ppm,  $0.06 \pm 0.001$  to  $0.13 \pm 0.006$  ppm, and  $0.03 \pm 0.001$  to  $0.11 \pm 0.005$  ppm respectively. Health risk assessment of these concentrations shows that all the values are below 1 and hence can only pose little or naught non-cancer/cancer risk to the consumers. However, people need to take caveat in consumption of herbal stuffs and agencies need to routinely check for quality of these stuffs to safeguard public health.

Keywords: Herbal snuff, lead, cadmium, chromium, risk, plant, food

# Introduction

Humans and other biota evolved in an ecosystem along with abiotic components consisting of metals (including heavy metals. Some of the metals are categorical examples of inorganic entities essential to the life of plants and animals in the ecosystem. Parable, Zn, Cu, Se, Fe, are (heavy) metals required by plants, and animals (including humans) for health, growth, and development (Iwuoha et al., 2013; Mafuyai et al., 2020). They are essential metals. The presence of useful (essential) metals in the environment encourage their uptake by plants from soil, and water more especially, for performing physiological roles, and other functions (Umar et al., 2023). Therewith, humans depend on plants for various purposes. Humans need plants to obtain food that provides nutrients (carbohydrates, lipids, vitamins, proteins, water, and minerals including metals as well, for instance) (Hashim et al., 2017; Horowilz et al., 2023). And on another basis, humans need plant-based products as source of medicines or bioactive substances that alter the human biological system in many respects such as physiological, morphological, etc. (Balamurugan et al., 2019; Sarkingobir et al., 2023). Current world spectrum characterized with a mixture of challenges affecting human existence, such as rising cost of living, rising rate of chronic diseases, rising rate of antibiotic resistance, and other misadventures; especially in developing countries like Nigeria, where healthcare is still at the verge of developing (Tukur et al., 2023). People in Nigerian rural areas are battling with poor health services, have to rely on alternative or traditional therapy (Muhammad et al., 2021; Sarkingobir et al., 2022).

Globally, the role of herbs/ phytochemicals in prevention and curing of diseases cannot be overemphasized. Actively, more than 2/3 of the world inhabitants pay much attention to herbal therapy for their healthcare needs because of properties such as lesser side effects, cost-effectiveness, and efficiency (Quds *et al.*, 2021; Al-thani *et al.*, 2023). There is a growing increase in utilization of herbs/ plant substances for various therapeutic applications especially in rural areas due to urbanization, and globalization among other reasons (Benson et al., 2017; Sarkingobir *et al.*, 2022). Many people believed in therapy with the application of herbs/plants and advocate on the cause parading the effectiveness, safety, cheapness, and accessibility of these products relying on empirical facts sand natural origin (Saeed *et al.*, 2011; Duburska *et al.*, 2022).

Certainly, worldwide, about 300 million people or more are using snuff products, in every ten adults, at least one is using snuff products in low-income countries (Salifu et al., 2023). It was reported

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that, from 113 countries, use of snuff products has led to about six million disability adjusted life years (DALYs) and spurred about 266, 592 cancers and heart disease and in turn leading to deaths (Salifu *et al.*, 2023). Snuff is a product made after pulverizing ingredients from herbs or plants (such as tobacco), other ingredients may be added deliberately or through contaminations (Muhammad *et al.*, 2021; Ajayi et al., 2023). Snuffing has been utilized as an alternative therapy for cold, pain, aches, grief, tiredness, etc. The utilizations of snuffing have been linked to effects such as cardiovascular diseases, and cancers (Salifu *et al.*, 2023). Prabahar (2012) in an Indian study shows that, tobacco-based snuffs contain heavy metals levels that may pose health risks. Owusu-Asante *et al.* (2022) determined levels of arsenic, cadmium, and lead in snuff in Shanti region, Ghana. The leaves were potentially toxic to humans. Investigation of chromium and cadmium in cigarette-based snuff shows potentially toxic levels in Rivers state, Nigeria (Ndokiari *et al.*, 2021). In a study in Ghana, consisting of 272 respondents, there was high intake of snuffs among the participants coupled with high lack of knowledge about hazards of snuff (Salifu et al., 2023). Ajayi et al. (2023) in a study in Ebonyi Nigeria analyzing snuffs for metals, show that, exposure to these snuffs may cause non-cancer health risks. Muhammad et al. (2021) reported in their experimental study in Jos that, certain snuff products cause oxidative stress in rat brain.

However, due to the growing concern about the rise in environmental pollution, chemicals including essential and non-essential heavy metals (such as lead, cadmium, and chromium) find ways into soil, water, air, food, and other environmental components and it is now imperative to monitor lead, chromium, and cadmium in herbal snuffs (Sarkingobir *et al.*, 2023). More prominently, heavy metals present in water and soil are taken up along with minerals and other nutrients (despite the strategies for abating pollutants in plants) for upward deposition in plant parts (Dahlawi et al., 2021; Quds et al., 2021; Ngumah *et al.*, 2022).

Consequently, when these plants are utilized as food or any other thing there is every possibility that the embedded heavy metals could be shuttled along the food chain and harm humans when certain exposure is ensured. Parable lead is a forefront toxic metal, capable of eliciting high blood pressure, and effects kidney and brain at slightest concentration. Cadmium too at low levels can elicit cardiac and kidney disorders. Similarly, chromium is toxic and is associated with human cancer (Duruibe *et al.*, 2007; Saeed *et al.*, 2011; Benson et al., 2017; Murphy et al., 2021). However, there is information scarcity about the levels and types of contaminants in herbal snuff being taken in Nigeria and Sokoto in particular. Therefore, the objective of this paper was to assess cadmium, chromium, and lead available in herbal snuff consumed in Sokoto, Nigeria and estimate human health risk.

# Materials And Methods Study area

The study was carried out in Sokoto state, Nigeria. Sokoto State is located in the Northwest Zone of Nigeria between longitude 11" 30–13" 50 and latitude 4"–6". It borders Niger Republic to the north and Benin Republic to the northwest, Kebbi State to south and Zamfara State to the east. It has a land mass area of about 32,000 km<sup>2</sup> and consists of 23 local government areas and 244 political wards. The population is predominantly rural, Muslim and consists almost entirely of Hausa/Fulani ethnic groups.

# Elemental and data analysis

Herbal snuffs are widely seen in Sokoto city. They are powdered materials (usually of plant origin) stocked in small containers that are snuffed by users to get some feelings of therapy on various issues affecting them. The name of each herbal powdered snuff is denoted mostly by name of its company or manufacturer which is boldly written on the containers. Since the snuffs are from different companies/manufacturers they may differ in effect and compositions, likewise their price and consumer preferences. The six different herbal snuffs namely, Hajiya Aisha, Hajiya safiya, Dr Lambo and others (some of them were shown in Figures 2-4) were purchased from Sokoto Market, Sokoto City, Sokoto State, Nigeria. The determination of heavy metals (lead, chromium, and cadmium) was performed by the methods of Association of Analytical Chemists (AOAC) described in Tukur et al., (2023).

# **Estimation of Human Health Risk Assessment**

Human Health risk was calculated using three different equations shown in this section. **CDI**=CP×IR×EF×ED/Bw×AT (mg/kg/day). Where, CDI= Chronic Daily Intake, CP= concentration of

metal in herbal snuff, IR=Ingestion Rate=1, EF= Frequency of Exposure=90 days, ED=Exposure Duration=30 days, Bw=weight=70 kg, AT= 2700 days.

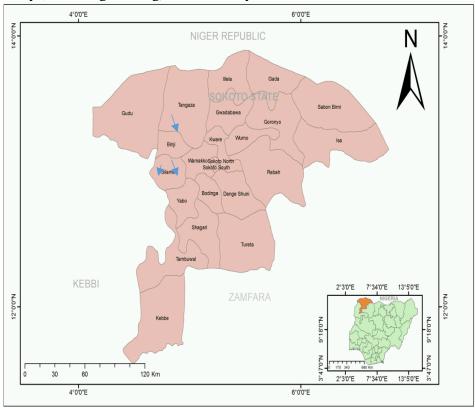


Figure 1. Map of Sokoto State, Nigeria, (Hamza et al. 2023).



# Hazard Quotient= CDI/RfD

Where, RfD= Chronic Oral Reference Dose, lead=3.5, chromium=3.0, cadmium=5.0. (Olagunju et al., 2020; Tschinkel et al., 2020).

**Hazard Index** (HI)= Summation of hazard quotient of Pb, Cd, and Cr= HQ  $_{cd}$  + HQ  $_{Cr}$ + HQ  $_{Pb}$  **EDI**= C×QMC/bw

EDI= Estimated daily intake, C= concentration of metal, QMC= 0.00788 g, quantity of snuff taken every day approximately from a review, bw= weight, it is 70 kg for adult.

# Statistical analysis

The descriptive statistics and one-way analysis of variance (ANOVA) were carried out at (p<0.05) significance level using Microsoft excel version 7.

# **Results and Discussion**

The results for this study were shown in Tables 1-5.

**Table 1.** Showing concentrations of lead, cadmium, and chromium metals assessed in some herbal snuffs collected from Sokoto, Nigeria

Type of herbal snuff	Lead (ppm)	Cadmium (ppm)	Chromium (ppm)
1=Snuff herbal powder	$0.03 \pm 0.001$	$0.08 \pm 0.0002$	$0.08 \pm 0.001$
2= Hajiya Ayisha snuff AK 47	$0.03 \pm 0.001$	$0.07 \pm 0.001$	$0.06 \pm 0.003$
3= Dr Lambo Special Sundu	$0.03 \pm 0.001$	$0.07 \pm 0.001$	$0.06 \pm 0.003$
4= Hajiya Ayisha snuff AK47 Blue Cover	$0.09 \pm 0.004$	$0.11 \pm 0.001$	$0.10 \pm 0.001$
5= Dr Lambo Herbal Powder	$0.11 \pm 0.005$	$0.12 \pm 0.005$	$0.04 \pm 0.001$
6= AK 47	$0.06 \pm 0.0001$	$0.13 \pm 0.005$	$0.13 \pm 0.006$

Keys: Values are expressed as mean  $\pm$  standard deviation

**Table 2**. CDI pertaining concentrations of lead, cadmium, and chromium metals assessed in some herbal snuffs collected from Sokoto, Nigeria

Snuff type	Cadmium	Chromium	Lead
1=Snuff herbal powder	0.001143	0.001143	0.00042857
2= Hajiya Ayisha snuff AK 47	0.001	0.0008571	0.00042857
3= Dr Lambo Special Sundu	0.001	0.0008571	0.00042857
4= Hajiya Ayisha snuff AK47 Blue Cover	0.001571	0.0014286	0.00128571
5= Dr Lambo Herbal Powder	0.0017143	0.0005714	0.001571
6= AK 47	0.0018571	0.0018571	0.0008571

**Table 3.** HQ pertaining concentrations of lead, cadmium, and chromium metals assessed in some herbal snuffs collected from Sokoto, Nigeria

Snuff type	Cadmium	Chromium	Lead
1=Snuff herbal powder	0.0002286	0.0004767	0.00012245
2= Hajiya Ayisha snuff AK 47	0.0002000	0.000381	0.00012245
3= Dr Lambo Special Sundu	0.0002000	0.000381	0.00012245
4= Hajiya Ayisha snuff AK47 Blue Cover	0.00031420	0.0004762	0.00036735
5= Dr Lambo Herbal Powder	0.00034286	0.000190467	0.00036735
6= AK 47	0.00185710	0.00190467	0.00024489

**Table 4**. Showing Hazard Index (HI) of concentrations of lead, cadmium, and chromium metals assessed in some herbal snuffs collected from Sokoto, Nigeria

Type of herbal snuff	Hazard Index (HI)
1	0.00511805
2	0.00070345
3	0.0052345
4	0.0020149
5	0.004206827
6	0.00621067

**Table 5**. Revealing the EDI pertaining concentrations of lead, cadmium, and chromium metals assessed in some herbal snuffs collected from Sokoto, Nigeria

Type of snuff		Cd	Pb
	1	2.10133333333E-5	0.00000078
	2	1.838666666E-5	0.00000078
	3	6.9869333333E-6	0.00000078
	4	6.9869333333E-6	0.000234
	5	2.8893333333E-5	2.889333333333E-5

6 0.000312 0.00001576

Table 1 shows the assessed concentrations of herbal snuff Cd, Cr, and Pb in some common herbal snuff consumed in Sokoto, Nigeria. The ranges of Cd, Cr, and Pb respectively are as follows: 0.07± 0.001 to  $0.13 \pm 0.005$  ppm,  $0.06 \pm 0.001$  to  $0.13 \pm 0.006$  ppm, and  $0.03 \pm 0.001$  to  $0.11 \pm 0.005$  ppm respectively. Nowadays, there is an increasing trend of utilization of herbal/traditional therapy across the various parts of the world, let alone in developing countries like Nigeria (Muhammad et al., 2021; Salifu et al., 2023). This trend could be attributed to the increase in burden of diseases, poor healthcare systems, poverty, and antibiotic resistance among others. Thus, it is imperative to perform assessment of quality of herbal stuffs; therewith, the need to assess heavy metals such as Cd, Cr, and Pb because they are capable of leading to toxicity in humans at any slight concentration (Ouds et al., 2021; Al-Thani et al., 2023. And the widespread pollution to anthropogenic activities that easily spoil food or other products (Olagunju et al., 2020; Labbo et al., 2021). Nevertheless, the non-essential heavy metals Cd, Cr, and Pb assessed in herbal stuff in Sokoto (as shown in Table 1) in 72.2% of the samples are within safe levels reported elsewhere. Parable, Pb determined in snuff (Table 1) is less than limit set by WHO (<10mg/kg), 250ug/day limit set by the Joint FAO/ WHO Expert Committee (JECFA), and 20ug/day limit set by American National Standards Institute (ANSI) (Mathew et al., 2021). This was in consonant with a survey of herbal stuffs in Pakistan that shows lead within permissible limit (Quds et al., 2021).

Cd concentrations  $(0.07\pm0.001\ to\ 0.13\pm00.005\ ppm)$  and Cr  $(0.06\pm0.001\ to\ 0.13\pm0.006ppm)$  shown in Table 1 depicting levels of non-essential heavy metals in snuff consumed in Sokoto are evident. The WHO permissible limit for Cd 0.3ppm conflicted with what was found in herbal varieties examined in this work. This contradicts a survey of Cd stuffs in Pakistan that shows Cd below the WHO limits (Quds et al., 2021). The presence of Cd in excess is either due to plants being cultivated in polluted soils, contamination during production, and leaching chemicals through from the containers (Balalimood et al., 2021; Mathew et al., 2021; Quds et al., 2021; Umar et al., 2023). Cd metal have the potential to affect respiratory system and spur cardiac and kidney disorders (Quds et al., 2021; Al-Thani et al., 2023). Therefore, there is need to routinely assess this metal in herbal medicines to safeguard public health. However, another Recommended Daily Limit (RDL) for cadmium reported in Saeed et al., (2011) of 70 ug was higher than the cadmium found in this study (Table 1). Additionally, another potentially toxic metal is chromium. Chromium concentrations when compared to the WHO permissible limit (30-35 ppm for adults and 11-25 ppm for children) shows a lower trend and was in conflict with results from Pakistani branded herbal medicines that contain higher Cr in 96% of the stuff studied (Saeed et al., 2011).

Moreover, since heavy metal consumption in foods and other sources are issues of concern, further assessment of the values obtained in Table 1 is imperative. The subjection of the results (in Table 1) to health risk assessment (HRA) was ensured. Table 2 shows the CDI for Cd, Cr, and Pb for snuff consumed in Sokoto, Nigeria; and fortunately, all the values are below 1. Thus, the population exposed to the snuff may not be at risk due to the metals in question with the assumption that consumers take in 1g/day (Mafuyai et al., 2020), further consumption above 1g could put the consumers at risk (Njoga et al., 2021. according to Table 3, HQ of Cd, Cr, and Pb in snuff was unveiled and all the values are below 1; therefore, it might not cause non-carcinogenic adverse effects on consumers using the observed concentrations (Table 1) (Njoga et al., 2021).

Similarly, Hazard Index (HI) for Cr, Pb, and Cd shows all values are below 1 for all the stuffs (Table 4). Thus, all the mixture of Cd, Pb, and Cr may not pose harm to the consumers at observed concentrations or may only lead to little harm (if there is any to the users) (Njoga et al., 2021). EDI, Estimated Daily Intake (Table 5) shows the amount that can be taken daily without expecting much risk and therewith all the values are below 1(Njoga et al., 2021). It is indeed imperative to assess non-cancer at these concentrations. Thus, the risk estimated for Cd, Pb using the observed concentrations (in Table 1) was shown in Table 5 and was fortunately lower than 1 and therefore there may be minor chance for the observed concentrations (Table 1) to elicit non-cancer risks (Njoga et al., 2021). Still people need to take care of when dealing with foods, and herbal stuffs because of possibility of pollution and agencies and scholars need to observe these herbal stuffs routinely before allowing public exposure.

# Conclusion

Herbal stuffs are getting popular world over, but there is concern about possible pollution, for instance with heavy metals. Thus, quality is routinely checked regarding heavy metals and other pollutants in herbal stuffs. This study reveals that health risk assessment of lead, cadmium and chromium concentrations divulge values that are below 1 and hence can only pose little or naught non-cancer/cancer risk to the consumers. However, people need to take caveat in consumption of herbal stuffs and agencies need to routinely check for quality of these stuffs to safeguard public health.

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